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FIXATION OF MOLECULAR NITROGEN BY THE ACTION OF ULTRASONIC WAVES
WITH THE FORMATION OF BIOLOGICALLY IMPORTANT SUBSTANCES

- USSR -

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FIXATION OF MOLECULAR NITROGEN BY THE ACTION OF ULTRASONIC WAVES
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[Following is a translation of an article by A. V. Sokol'skaya and I. Ye. El'piner in the Russian-language periodical Akusticheskiy Zhurnal (Acoustics Journal), Moscow, Vol. VI, No. 2, 1960, pages 263-264.]

In previous reports [refs. 1, 2] it was shown that, under the action of ultrasonic waves in hydrogen- and nitrogen-saturated water, ammonia is synthesized; and in the presence of hydrogen, nitrogen and carbon dioxide or methane, hydrocyanic acid and formaldehyde are synthesized. In other words, in water, in the presence of oxygen, nitrogen is fixed with the formation of products useful for further synthesis of biologically important substances.

Table 1

The Amount of Nitrogen-Containing Organic Compounds (Oximes)
Formed as a Result of the Fixation of Molecular Nitrogen by
Aliphatic Acids in an Ultrasonic Wave Field

Sonically Irradiated Substance	Concentration of Irradiated Substance, %	Amount of Oxime* Formed in γ ($\gamma = 10^6$ g) per 1 ml of Solution Irradiated in the Presence of /See Note/	Nitrogen
Citric acid ($\text{COOH}-\text{CH}_2-\text{C}(\text{OH})\text{COOH}-\text{CH}_2-\text{COOH}$)	1.0	0.2	1.6
Malic acid ($\text{COOH}-\text{CH}(\text{OH})-\text{CH}_2-\text{COOH}$)	1.0	0.1	2.8
Fumaric acid ($\text{COOH}-\text{CH}=\text{CH}-\text{COOH}$)	0.4	0.5	2.7
Succinic acid ($\text{COOH}-\text{CH}_2-\text{CH}_2-\text{COOH}$)	1.0	0.6	3.0

<u>Sonically Irradiated Substance</u>	<u>Concentration of Irradiated Substance, %</u>	Amount of Oxime* Formed in γ ($\gamma = 10^6$ g) per 1 ml of Solution Irradiated in the Presence of /See Note/ <u>Air</u> <u>Nitrogen</u>	
α -Ketoglutaric acid (COOH-CH ₂ -CH ₂ -CO- COOH)	1.0	0	0
Distilled water	--	0	0
Sulfuric acid	0.01 N	0	0

* On the basis of N_2O_3 .

Note: The figure 10⁶, in the original, should obviously be 10⁻⁶.]

Actually our studies have shown that nitrogen activated or reduced by ultrasound is fixed by a number of organic compounds with the formation of substances which play an important role in the metabolism of living and growing cells. We are considering the possible formation of the amino acids which constitute protein particles. In our work we have used aqueous solutions of organic acids, saturated with nitrogen or mixtures of nitrogen with oxygen, argon, or helium. The gases (nitrogen, argon or helium) were rid of oxygen by passing them several times through absorbers containing alkaline solution of pyrogallol.

The aqueous solutions (10 ml) were sonically irradiated at 560 kilohertz with an intensity of 12 watts/cm² and with the irradiated medium at a temperature of 25-30°. After two hours' sonic irradiation of an aqueous solution saturated with molecular nitrogen and containing citric, succinic, fumaric and other aliphatic acids, the corresponding oximes R:NOH were formed (where R is the organic acid radical). To identify the oximes, we used a method which permits the quantitative determination of nitrite formed as a result of the splitting off of hydroxylamine from the organic molecule being studied, with the former being subsequently oxidized to nitrogen oxides.

As shown in Table 1, nitrogen fixation by organic acids with the formation of oximes is mostly expressed during the sonic irradiation of dibasic organic acids (succinic, malic, and fumaric acids). It is interesting that the presence of air depresses the process of nitrogen fixation by dibasic acids. In the presence of air (oxygen), succinic acid fixes nitrogen 5 times less than in the presence of nitrogen alone. Malic acid almost does not combine with nitrogen in air, but its fixation of nitrogen is 20

times greater in pure nitrogen. The depressing effect of molecular oxygen on nitrogen fixation by aliphatic acids is evident from the data given in Table 2.

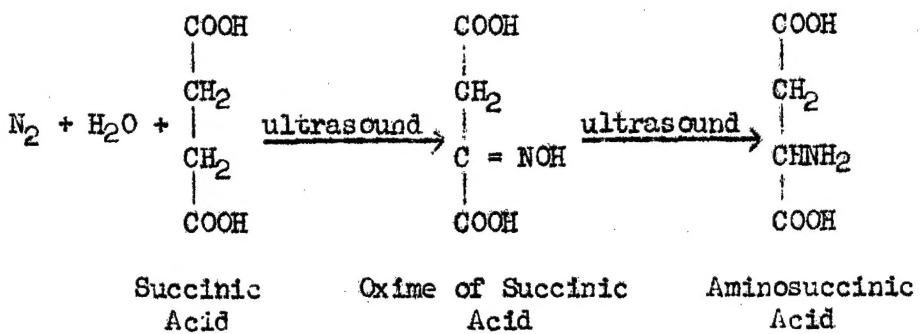
Table 2

The Amount of Oxime of Succinic Acid Formed Upon Sonic Irradiation
in the Presence of Different Gases
(Sonic Irradiation Period, 2 Hours)

<u>Solution Studied</u>	Amount of Oxime Formed, in γ ($\gamma = 10^{-6}$ g) per 1 ml* of Succinic Acid, Irradiated in the Presence of				
	N	N and A	N and He	N and O	Air
1% aq. sol. succinic acid	3.3	2.5	1.9	0	0.6
"	3.0	2.4	1.9	0	0.3

* On the basis of N_2O_3 .

As shown in the table, the greatest amount of oximes of succinic acid were obtained by sonically irradiating the solution in the presence of nitrogen alone, a somewhat lesser amount being found in a mixture of nitrogen with argon or helium. Nitrogen fixation by succinic acid stops altogether if the solution is irradiated in a mixture of nitrogen and oxygen. It was further shown that, with longer periods of sonic irradiation (8-10 hours), amino acids were probably formed in the aqueous solution (of succinic acid in the presence of nitrogen). This was verified by the following method: After final sonic irradiation, the solution was vaporized in vacuo to distill off volatile amines and organic acids. The concentrate obtained was subjected to paper chromatography. As a solvent for chromatographic analysis, a mixture of butyl alcohol, acetic acid and water (4:1:5) was used. A 0.02% solution of ninhydrin in acetone was used to develop the chromatogram. On the chromatograms made in this way, we invariably discovered several spots. The spots had the characteristic rose-red color of amino acids. Identification of the spots showed that they belonged to aminosuccinic acid, 2-amino-propanoic acid, and several other acids. The phases of the reaction may be represented schematically as follows:



In this way, fixation of molecular nitrogen by organic compounds with the formation of amino acids is accomplished at near room temperature without biocatalysts (ferments) if aqueous solutions of these compounds are subjected to the action of ultrasonic waves in the presence of gaseous nitrogen. This opens new possibilities in the field of ultrasonic chemistry and biology.

References

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2. El'piner, I. Ye., Sokol'skaya, A. V., "Synthesis of Substances in Water Saturated With Gases, Under a Reduced Atmosphere, by the Action of Ultrasonic Waves," Dokl. AN SSSR /Reports of the Academy of Sciences USSR/, 1958, 119, 6, 1180-1182.